



The Effects of Fluvalinate and Coumaphos on Honey Bees in Two Commercial Queen Rearing Operations

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Abstract

We conducted research on the potential impacts of fluvalinate and coumaphos on queen viability and health. Queens were reared in colonies that had been treated with differing amounts of both fluvalinate and coumaphos. Pre- and post-treatment samples of both wax and bees were collected from all of the colonies and analyzed for total concentrations of fluvalinate and coumaphos. All queens were measured for queen weight, ovarian weight, and number of sperm in the spermathecae. We measured these characteristics to determine if the treatments have any effect on queen development and mating above the normal variation seen among queens for these characteristics. The queens treated with high doses of fluvalinate (8 Apistan® strips) weighed significantly less than low dose (2 Apistan® strips) or control queens, but otherwise appeared to develop normally. The highest fluvalinate concentrations were observed in the wax and queen cells of the high dose group. The developing queens treated with varying levels of coumaphos suffered a high mortality rate. In general, acceptance of coumaphos exposed queen cells was very low. Successful production of queens was difficult when coumaphos was present in the starter colonies for any extended period of time. Many attempts were made to rear queens, using various amounts of coumaphos for varying time periods, before queens could be successfully produced. High mortality of larvae was noted in colonies that contained as little as one CheckMite™ strip of coumaphos for more than 24 hours. Several of the queens showed sub-lethal effects from the coumaphos, including physical abnormalities and atypical behavior. The queens exposed to coumaphos weighed significantly less and had lower ovary weights than the control group queens. The highest coumaphos concentrations were observed in the queen cells and wax of the high dose groups.

Introduction

While U.S. beekeepers produce in excess of 200 million pounds of honey annually, the principle benefit of beekeeping is pollination of food crops. A recent study by Cornell University researchers valued the benefit of honey bee pollination to U.S. agriculture at \$14.6 billion annually. Wild pollinating insects contribute to crop pollination, but in recent years their abundance has declined in many areas due to urban expansion, monocultural cropping practices, introduced bee parasites, loss of nesting habitat, reduction in forage plants, and pesticide use. As a result, managed pollinating insects are increasingly important to farmers, gardeners, and orchardists.

The beekeeping industry has entered a new era with the recent widespread use of miticides to treat the parasitic mite, *Varroa jacobsoni*. For the last ten years, the synthetic pyrethroid, fluvalinate (Apistan®), has been used very successfully to treat the mites. However, in the last two years, the mites have developed resistance to this miticide. To relieve the crisis that emerged with the resistant mites, many states obtained Section 18 (EPA) approval for use of the organophosphate, coumaphos. The coumaphos and fluvalinate impregnated strips used in the U.S. are readily absorbed and accumulated into beeswax. There has been concern about the lack of research that has been conducted on the possible ill-effects of miticides to bees.

In recent years, some beekeepers have had considerable problems with queen loss and queen supersede. The possibility has been raised that the increased use of miticides may be having a negative impact on queen health and viability. In our experiments, we researched the potential impacts of fluvalinate and coumaphos on queen viability and health.

Fluvalinate-California Study

This experiment was conducted in San Diego during July 2000. We grafted queen bee cells into honey bee colonies that were then treated in one of three ways:
•LOW DOSE GROUP- 2 Apistan® strips attached lengthwise to cell bars
•HIGH DOSE GROUP- 8 Apistan® strips: 4 attached to cell bars and 4 hanging
•CONTROL- plastic dummy strip
Bee and wax samples were collected at the beginning and end of the experiment and analyzed for fluvalinate using Gas Chromatography with Electron Capture Detection (Table 1). Queen cells were placed into mating nucs containing approximately 1/2 lb of bees. The Low Dose Group received 1/2 strip per nuc and the High Dose Group received 1 strip per nuc. In September, the queens were collected and sent to University of Minnesota and examined for queen weight, ovarian weight, and number of sperm in spermatheca (Table 2).

Table 1. Fluvalinate Concentrations in the Bees, Wax, and Queen Cells

Sample Collected	Residue Concentration (ppb/kg)	
	Fluvalinate	Final Concentration
	0.001	0.001
	Detection Limit < 0.25	Detection Limit < 0.25
Bees		
2-strip Group	0.13	0.11
8-strip Group	1.35	0.20
Control Group		
Wax		
2-strip Group		Below Detection Limit
8-strip Group	4.27	2.58
Control Group	0.40	1.05
Queen Cells		
2-strip Group	N/A	5.70
8-strip Group	N/A	2.28
Control Group	N/A	6.74

Table 2. Results of Queen Viability Assessments Using Queens from the Three Fluvalinate Groups

Treatment	Mean Queen Weight (g)	Mean Ovary Weight (g)	Mean Number of Sperm
Control	0.197	0.046	4,058,708
(n = 20)	±0.003	±0.016	±1,077,199
2-strip	0.200	0.050	4,064,739
(n = 10)	±0.003	±0.008	±1,425,377
8-strip	0.181	0.038	3,628,752
(n = 12)	±0.004	±0.004	±1,042,489

Methods/Results

Coumaphos-California Study

This experiment was conducted near San Diego starting in July 2000. We made several attempts at finding sublethal doses to developing queen larvae. We first used treatments consisting of 1 to 4 strips of CheckMite +™ attached to cell bars, but all the queens died after the cells were sealed. We also attempted to treat the developing larvae for a 24-hour period, but all queens died, including one that was deformed (beet antennae and uncoordinated movements). Finally, we successfully raised queens using the following treatments:
•LOW DOSE GROUP- 1/4 strip, as above
•HIGH DOSE GROUP- 1/2 CheckMite +™ strip on the cell bar
•CONTROL- plastic dummy strip
Even at these treatment levels, we still discovered several queen deformities of antennae and hind legs. At the end of the experiment, concentrations of coumaphos within treatment groups were analyzed using composite samples of wax, bees, and queen cells using Gas Chromatography Thermionic Specific Detector (Table 3). Surviving queens were sent to University of Minnesota and measured for queen weight, ovarian weight, and number of sperm in spermatheca (Table 4).



Table 3. Coumaphos Concentrations from California Study

Sample Collected	Residue Concentration (ppb/kg)	
	Coumaphos	Final Concentration
	0.001	0.001
	Detection Limit < 0.25	Detection Limit < 0.25
Bees		
1/4-strip		Below Detection Limit
1/2-strip	1.54	0.47
Control		Below Detection Limit
Wax		
1/4-strip	0.30	40.26
1/2-strip	0.26	120.00
Control		0.47
Queen Cells		
1/4-strip	N/A	181.00
1/2-strip	N/A	227.00
Control	N/A	Below Detection Limit

Table 4. Results of Queen Viability Assessments Using Queens from the California Coumaphos Groups

Treatment	Mean Queen Weight (g)	Mean Ovary Weight (g)	Mean Number of Sperm
1/4-strip	0.189	0.035	3,847,917
(n = 20)	±0.004	±0.004	±1,223,937
1/2-strip	0.191	0.036	3,297,140
(n = 20)	±0.003	±0.005	±1,060,171
Control	0.197	0.040	4,048,750
(n = 20)	±0.003	±0.004	±1,077,199

Coumaphos-Texas Study

A similar study was conducted in Navasota starting in May 2000. Queen cells were grafted into starter colony (10 lbs bees). After many trials to find a non-lethal dose, and many dead queens, viable queens emerged with the following doses:
•2 CheckMite +™ strips attached lengthwise to cell bars (1/2 strip per bar)
•2 CheckMite +™ strips hanging from cell bars, adjacent to cells
•2 CheckMite +™ strips hanging but not adjacent cells
•Control - plastic dummy strip
Concentrations of coumaphos within treatment groups were analyzed (Table 5), and all queens were sent to University of Minnesota (Table 6).

Table 5. Coumaphos Concentrations from Texas Study

Sample Collected	Concentration (ppb/kg)	
	Coumaphos	Detection Limit < 0.25
Bees		
2-strip Attached	6.56	
2-strip Adjacent	23.26	
2-strip Not Adjacent	9.95	
Control	0.27	
Wax		
2-strip Attached	12.00	
2-strip Adjacent	23.12	
2-strip Not Adjacent	12.85	
Control		Below Detection Limit
Queen Cells		
2-strip Attached	91.93	
2-strip Adjacent	28.17	
2-strip Not Adjacent	0.31	

Table 6. Results of Queen Viability Assessments Using Queens from the Texas Coumaphos Groups

Treatment	Mean Queen Weight (g)	Mean Ovary Weight (g)	Mean Number of Sperm
2-strip Attached	0.1708	0.0404	4,071,620
(n = 6)	±0.0035	±0.0018	±1,040,390
2-strip Adjacent	0.1800	0.0408	4,064,676
(n = 7)	±0.0042	±0.0053	±1,778,544
2-strip Not Adjacent	0.1978	0.0417	3,630,802
(n = 6)	±0.0033	±0.0015	±1,767,700
Control	0.2123	0.0479	4,060,001
(n = 7)	±0.0049	±0.0085	±1,178,911

Statistical Results

Fluvalinate-California Study

Our statistical analysis indicated that queens reared in high dose (8 strips throughout rearing, 1 strip in mating nuc) weighed significantly less ($p = 0.01$) than low dose and control queens. There was no significant difference between the ovary weights ($p = 0.27$) or the mean number of sperm ($p = 0.08$).

Coumaphos-California Study

Our statistical analysis indicated that all treated queens weighed significantly less ($p = 0.002$) and had lower ovary weights ($p = 0.001$) and lower sperm counts ($p = 0.001$) than control queens.

Coumaphos-Texas Study

Statistical analysis indicated that queen weights ($p = 0.004$) and ovary weights ($p = 0.004$) of "2-strips attached" group were significantly less than control queens.



Conclusions

Fluvalinate

- Fluvalinate treated queens appeared to develop normally.
- The concentrations of fluvalinate in the bees were variable. The levels were highest in the wax and queen cells.
- High doses of fluvalinate during queen development may result in lower queen weights.

Coumaphos

- High doses of coumaphos in cell starters, or coumaphos touching queen cells, will likely kill queen larvae.
- Queens that do develop may have deformities.
- Queens that do develop may be lighter and have reduced ovary weights. Sperm count differences may depend on drone availability.
- Currently, the EPA Tolerance Level for beeswax is 100 ppm. The concentrations observed in the California Study queen cells and high wax group were all above 100 ppm. The Texas Study concentrations were below 100 ppm.
- Reduced queen and ovary weights are occurring at concentrations below the EPA Tolerance Level.



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